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Development of a chromosomal arm map for wheat based on RFLP markers *

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Summary. A chromosomal arm map has been developed for common wheat (*Triticum aestivum* L. em. Thell.) using aneuploid stocks to locate more than 800 restriction fragments corresponding to 210 low-copy DNA clones from barley cDNA, oat cDNA, and wheat genomic libraries. The number of restriction fragments per chromosome arm correlates moderately well with relative DNA content and length of somatic chromosomes. The chromosomal arm locations of loci detected with 6 different clones support an earlier hypothesis for the occurrence of a two-step translocation (4AL to 5AL, 5AL to 7BS, and 7BS to 4AL) in the ancestral wheat genomes. In addition, 1 clone revealed the presence of a 5AL segment translocated to 4AL. Anomalies in aneuploid stocks were also observed and can be explained by intrahomoeologous recombination and polymorphisms among the stocks. We view the development of this chromosomal arm map as a complement to, rather than as a substitute for, a conventional RFLP linkage map in wheat.

Key words: Aneuploid-chromosomal map-RFLPs-Translocation-*Triticum aestivum*

Introduction

The rationale and use of restriction fragment length polymorphism (RFLP) maps in crop improvement have been extensively reviewed (Helentjaris et al. 1985; Tanksley et al. 1989). RFLP maps of useful levels of saturation have been produced for several diploid crop species or their relatives including barley (Heun et al. 1991), lettuce (Landry et al. 1987), maize (Helentjaris 1987), potato (Bonierbale et al. 1988), rice (McCouch et al. 1988), soybean (Apuya et al. 1988), tomato (Bernatzky and Tanksley 1986), and *Triticum tauschii* (Gill et al. 1991). In cultivated bread wheat (*Triticum aestivum* L. em. Thell.), an allohexaploid (2n = 6x = 42), an RFLP map will be particularly useful as homoeoloci of each genome could be visualized.

The development and use of RFLP markers in wheat has been slowed because of limited polymorphism in this crop, (Chao et al. 1989; Kam-Morgan et al. 1989; Liu and Tsunewaki 1990; unpublished data), likely due to its relatively recent origin (Bell 1987). Bread wheat has three genomes and theoretically would require 3 times the number of polymorphic restriction fragments as a diploid to construct an RFLP linkage map of equal density. Despite these difficulties, there is a great interest and potential in applying RFLP technology to wheat improvement. Recently, an RFLP linkage map of homoeologous group 7 chromosomes of hexaploid wheat has been constructed (Chao et al. 1989).

One approach to facilitate the development and application of RFLPs in wheat is the construction of a chromosomal arm map. Wheat is uniquely suited for such a mapping strategy because of an abundance of aneuploid stocks (Sears 1954). Since the 1950s, breeders and geneticists have made use of these aneuploids to determine the chromosomal location of many genes (for review, see Milne and McIntosh 1990; Hart and Gale 1990). The objective of the research reported here was to locate a large number of DNA restriction fragments corresponding to single or low-copy clones to chromosome arms in wheat using aneuploids so that these clones may be applied to gene tagging, linkage and mapping of quantitative trait loci (QTL), cytogenetic manipulations, esti-

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mation of genetic distance, and genetic studies. This study also adds to our knowledge of the comparative organization of homoeologous chromosomes in wheat.

Material and methods

Genetic stocks and Southern hybridizations

Seeds of nullisomic-tetrasomic (NT) lines of 'Chinese Spring' wheat (Sears 1966) (complete except for 2A and 4B) and ditelosomics (DT) of 'Chinese Spring' (Sears and Sears 1978) (complete except for 2AL, 4AS, 5AS, 2BS, 4BL, 5BS, 5DS, and 7DL) were kindly provided by ER Sears, University of Missouri. An independently derived set of DTs of 'Chinese Spring' provided by Y. Ogihara, Kihara Institute, was also used for some probings. The designations of 4A and 4B used here are those agreed upon at the Seventh International Wheat Genetics Symposium held in 1988 at Cambridge, UK.

Leaf DNA was extracted from 3- to 4-week-old seedlings (bulk of four or five) by a procedure modified from McCouch et al. (1988) using an extraction buffer containing no urea or phenol (Tai and Tanksley 1990) and omitting the second DNA precipitation. DNA dissolved in TE buffer was digested with approximately 2 units of endonuclease/μg DNA in the presence of the appropriate buffer. Digestions proceeded for 16 h at 37 °C and were stopped by adding 10% w/v gel-loading buffer (blue-juice) (Heun et al. 1991). Digested DNA was loaded onto gels (approximately 20–25 μg per lane), electrophoresed, and transferred to Hybond N^+ membranes (Amersham, Arlington Heights, Ill) as described by Heun et al. (1991). Prehybridization of membranes, hybridization, washing, and exposure were as described by Heun et al. (1991) except that exposures of autoradiograms were for 1–7 days. Membranes were stripped by pouring near-boiling (96–99 °C) 0.5% SDS over them and agitating for approximately 30 s.

Clone selection and chromosome arm assignment

The clones used were from barley cDNA (BCD), oat cDNA (CDO), and wheat genomic (WG) libraries previously described by Heun et al. (1991). Wheat chromosome locations of the clones BG 131 and DG F15 from barley and *Triticum tauschii* genomic libraries, respectively, used previously in the construction of a barley RFLP map (Heun et al. 1991) are also reported here. Clones from each library were pre-screened by hybridization with membranes containing DNA of 'Chinese Spring' (the genetic background of the aneuploid stocks) digested with the restriction enzymes *Eco*RI, *Eco*RV, and *Dra*I. Low-copy clones (those hybridizing to 6 or fewer discernible fragments) were probed onto membranes containing DNA of NTs and DTs digested with one of the three restriction enzymes. Preference was given to clone/enzyme combinations that yielded 3 fragments (presumably 1 for each genome) of approximately equal hybridization intensity. Resulting films of the NT/DT probing were visually scored to identify fragments absent in any of the stocks. If a fragment was absent in a particular NT stock we inferred its location on the chromosome in the nullisomic condition. Concomitant presence of a double-dose fragment in the stocks tetrasomic for a particular chromosome was used as additional evidence for the proper localization of fragments except in the case of 2A and 4B. For these two chromosomes, clone assignment was based on the presence of double-dose fragments in the tretrasomic stock since nullisomics for these two chromosomes were not available. In the analysis of the DT's, a fragment absent in a stock indicated its presence on the opposing arm of that chromosome (i.e., a fragment absent in DT 1AL would indicate that fragment originated from the short arm of chromosome one). In those cases where a complete ditelosomic set was not available, the assignment of restriction fragments to chromosome arms was inferred based upon the presence of the fragment in the ditelosomic stock available. For each clone, fragments were classified as being major or minor according to their relative intensities. Minor fragments were those with an approximately 50% weaker autoradiographic signal than major fragments produced by the same clone. For most of the selected clone/enzyme combinations, 3 major fragments were observed, a varying number of minor fragments, and also faint fragments at our moderate stringency of $0.5 \times$ SSC at 65 °C. Fragments that accounted for less than approximately 5% of the total hybridization signal were not analyzed. Twenty-eight of the clones were mapped using more than one enzyme to confirm fragment locations when there were ambiguities. In those cases, only data from one enzyme was included when totalling the number of restriction fragments located.

Results and discussion

Distribution of mapped loci

A total of 804 restriction fragments corresponding to 210 clones were assigned to wheat chromosome arms (Table 1). Overall, 88% of all hybridizing fragments considered reproducible were located to a chromosome or chromosome arm. The fragments not disappearing in any of the aneuploid stocks are likely the result of co-migrating restriction fragments. All fragments from 116 out of the 210 clones were assigned to a chromosome arm. An example of a probing in which 3 fragments were assigned to each of the three homoeologous long arms of group 3 chromosomes is shown in Fig. 1.

The assigned loci from randomly chosen clones (198 out of 210) are not uniformly distributed on all chromosome arms (Fig. 2). Homoeologous group 6 chromosomes are the least populated, having only 61 loci (8.2% of the 745 fragments located from random clones) compared to 14.8%, 16.5%, 16.1%, 12.8%, 18.3%, and 13.4% for homoeologous groups 1, 2, 3, 4, 5, and 7 chromosomes, respectively. Heterogeneity of the BCD and CDO libraries for the total number of restriction fragments located to chromosomes was significant and greatest for groups 3 and 7 ($P < 0.05$) (Table 2). There were not enough wheat genomic clones located to make a valid comparison.

Correlations among the distribution of restriction fragments, somatic chromosome length (Gill 1987), and DNA content measured by densitometry readings of stained metaphase chromosomes of 'Chinese Spring' (Furuta et al. 1988) were calculated (Table 3). The distribution of restriction fragments assigned from both the BCD and CDO libraries correlated better with somatic chromosome length than with DNA content. These lower correlations compared to that between DNA content and chromosome length ($r = 0.906$) may be explained by

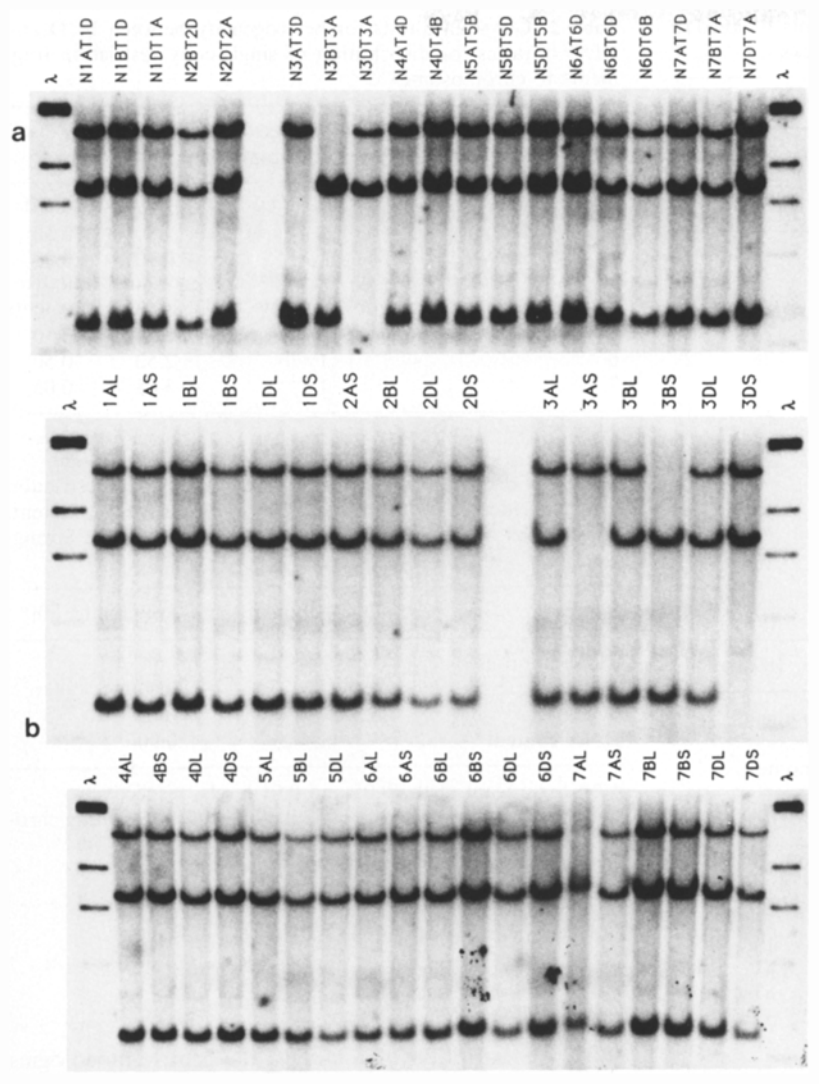

Fig. 1 a, b. Autoradiogram from clone BCD 1127 probed onto 'Chinese Spring' wheat aneuploids digested with *Eco*RV. λ indicates lambda DNA digested with *Hin*dIII and used as a size marker. **a** Result of probing onto nullisomic (*N*)-tetrasomics(*T*). Each of the three fragments corresponds to one of the genomes of the group 3 chromosomes. **b** Result of simultaneous probing onto ditelosomic stocks. The fragments were assigned to the long arms of group 3 chromosomes based on their absence in 3BS, 3AS, and 3DS

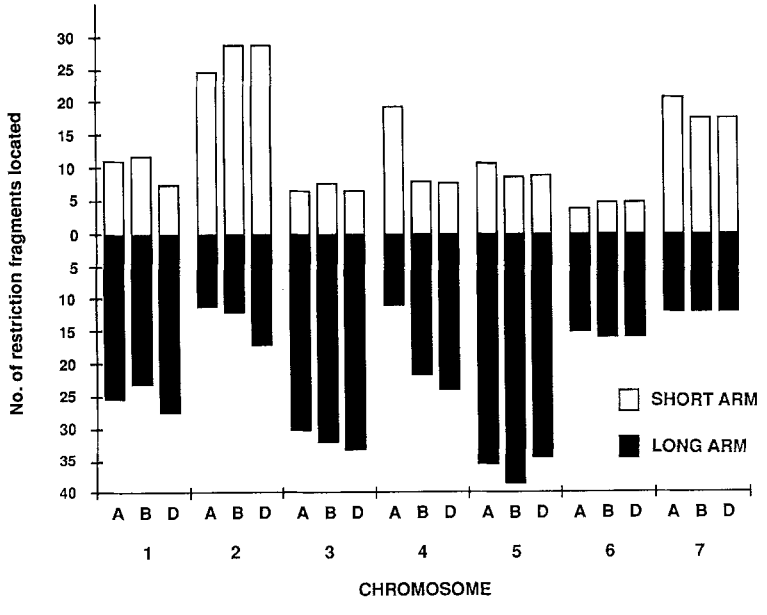

Fig. 2. Numbers of DNA restriction fragments from randomly chosen barley cDNA, oat cDNA, and wheat genomic clones located to individual chromosome arms using aneuploids of 'Chinese Spring' wheat. Fragments assigned to a chromosome group but not an arm are not included

Table 1. Number of clones and restriction fragments assigned to chromosome arms of wheat using aneuploid stocks

Fragments assigned to one homoeologous group	No. of clones	Number of fragments			
		Total	By genome		
			A	B	D
1L	18	53	18	17	18
1S	8	22	7	8	7
2L	11	42	13	13	16
2S	16	67	20	24	23
3L	23	86	29	28	29
3S	6	25	10	8	7
4L	16	48	16	15	17
4S	3	14	4	6	4
5L	22	78	25	28	25
5S	6	18	6	6	6
6L	11	37	12	13	12
6S	2	7	2	2	3
7L	10	32	10	10	12
7S	10	29	10	9	10
7	1	4	1	2	1
Total	163	562	183	189	190
Fragments assigned to more than homoeologous group	No. of clones	Number of fragments			
		Total	By genome		
			A	B	D
1L	13	31	11	9	11
1S	4	8	3	4	1
1	1	4	2	1	1
2L	4	6	0	1	5
2S	5	16	5	5	6
3L	9	24	6	9	9
3S	2	6	2	2	2
3	1	3	0	2	1
4L	12	22	8	7	7
4S	4	9	3	2	4
4	2	3	2	1	0
5L	18	40	14	14	12
5S	4	11	5	3	3
6L	5	11	3	3	5
6S	2	7	2	3	2
7L	4	7	3	3	1
7S	12	28	11	9	8
7	2	6	1	4	1
Total	47[a]	242	81	82	79

[a] Includes 34, 3, and 3 clones with fragments assigned two, three, and four homoeologous groups, respectively, and 7 clones that reveal translocations

either a general lack of coding sequences and/or the presence of a greater proportion of repeated DNA relative to other chromosomes for those regions under-represented on the arm map. Possible non-uniform placement of clones on this arm map should not diminish their usefulness since only those clones corresponding to low-copy sequences are useful for most applications.

Table 2. Chi-square tests for heterogeneity between BCD and CDO libraries for distribution of single-copy restriction fragments to chromosomes

Homoeologous chromosome groups compared	Degrees of freedom	χ^2	$P(<)$
ALL	6	23.77	0.001
1	1	1.86	0.50
2	1	0.90	0.90
3	1	7.74	0.01
4	1	1.63	0.50
5	1	2.34	0.50
6	1	2.52	0.50
7	1	3.85	0.05

Table 3. Correlation coefficients among number of restriction fragments located from BCD and CDO libraries, DNA content, and lengths of somatic chromosome arms of 'Chinese Spring' wheat[a]

Source	DNA content[b]	Somatic length[c]
Clone libraries		
BCD	0.440**	0.539**
CDO	0.321*	0.397**
DNA content[b]		0.906**

*,** Significant at the 0.05 and 0.01 levels, respectively

[a] Each correlation consists of 42 data points representing chromosome arms of each genome

[b] From Furuta et al. (1988)

[c] From Gill (1987)

Arm homoeologies

With the exception of 3 clones, the arm homoeologies revealed by probings of the aneuploid stocks agree with those previously deduced. Within chromosome groups, long arms were homoeologous to long arms and, consequently, short arms with short arms, with the exception of group 4 chromosomes in which 4AL = 4BS = 4DS and 4AS = 4BL = 4DL. These results are consistent with those reported by Hart (1973) based on the locations of isozymes of alcohol dehydrogenase and acid phosphatase, and later supported by the locations of isozymes of lipoxygenase (Hart and Langston 1977). The group 4 homoeologies can be explained by an inversion in chromosome 4A during the evolution of wheat.

The clones yielding exceptions to expected arm homoeologies were BCD 446 (fragments assigned to 1AL, 1BS, and 1DS) and two chromosome group 4 clones (BCD 1262 and CDO 669). The BCD 446 result may be due to the presence of a small pericentric inversion in 1A. Mistakes due to mislabelling or gel loading can be ruled out because of the results of CDO 580 (fragments assigned to 1AS, 1BS, and 1DS) using the same membrane. One group 4 exception, CDO 669 (fragments assigned to

4AL, 4AS, 4BL, and 4DS), could be explained by polymorphism among the aneuploid stocks since one of the two 4A-specific fragments is absent in DT4 AL while the other is at about twice the relative intensity of the same fragment in other stocks. The results of clone BCD 1262 (fragments assigned to 4AL, 4BL, 4DL) indicate that the pericentric inversion believed to have occurred in 4A did not involve the entire long arm.

Duplications

Thirty-four clones hybridized to multiple fragments on the same chromosome. This result might be attributed to intrachromosomal duplication of loci, and/or the presence of restriction sites within the chromosomal segment hybridizing to the clone. Forty clones hybridized to fragments on non-homoeologous chromosomes (excludes clones involved in translocations) and may represent interchromosomal duplications. For 34 of these 40 clones, only two chromosome groups were involved. In the majority of these cases, 3 or more major fragments were assigned to chromosomes in one homoeologous group and minor fragments were assigned to the other group. All fragments from all enzymes tested were assigned to a chromosome from 18 of the 40 clones revealing interchromosomal duplications. For the remainder of the clones, it is likely that additional fragments could be assigned to a chromosome if more enzymes were used. Group 1 chromosomes were involved in the most duplications (15 of 34 clones) compared to 6, 8, 10, 14, 5, and 10 for homoeologous group 2, 3, 4, 5, 6, and 7 chromosomes, respectively (Table 4). The three genomes (A, B, or D) were involved in about the same number of duplications.

Evidence for homoeologous recombination in N5BT5D

The 42 chromosomes of hexaploid wheat pair preferentially as 21 bivalents (7 per genome) due largely to the effect of the *ph* gene, located on chromosome 5BL, which suppresses homoeologous recombination (Okamato 1957; Riley and Chapman 1958). Other homoeologous pairing inhibitors/enhancers with lesser effects have been identified as well (see Gale and Miller 1987). We have detected homoeologous recombination between the short arms of chromosomes 2A and 2D in the nullisomic 5B, tetrasomic 5D (N5BT5D) stock. This putative recombination event(s) is evidenced by 6 clones (BCD 348, CDO 418, CDO 426, CDO 783, CDO 666, and CDO 981) assigned to homoeologous group 2 chromosomes. For these clones, the same molecular weight fragment(s) was absent from N2DT2A and N5BT5D and compensated for by the presence of the corresponding double-dose fragment(s) (Fig. 3). Mistakes due to mislabelling of stocks or loading of gels were ruled out since other clones probed to the same membranes had fragments missing in only the homoeologous group 2 or 5 stocks (data not shown).

Table 4. Number of clones detecting interchromosomal group duplication of loci for different combinations of chromosomes

Homoeologous group	Homoeologous group					
	2	3	4	5	6	7
1	1	5	3	2	0	4
2		1	2	2	0	0
3			0	1	1	0
4				4	0	1
5					2	3
6						2

A single fragment was absent in more than one lane within a set of NTs or DTs for 14 other clones as well; however, no compensating double-dose fragment was observed. This could be the result of homoeologous recombination due to the action of other pairing inhibitors/enhancers, insertion, deletion, or loss of a restriction site in which the size of the "new" fragment is less than 1 kb or more than 25 kb, the typical size range that can be detected in Southern hybridizations. For all cases in which the same fragment was absent in more than one lane with a set of NTs or DTs, the fragment(s) was assigned to only one chromosome arm based on the location of other fragments and assuming homoeology.

Evidence for translocations

Based on the pairing frequencies of chromosomes in 5B or 3D deficient lines (which enhances the frequency of homoeologous recombination) of 'Chinese Spring' wheat, a double translocation of 4AL to 5AL, 5AL to 7BS, and 7BS to 4AL has been proposed by Naranjo et al. (1987). This proposal is supported by the location of structural genes for the isozymes of β-amylase on chromosome arms 4BL, 4DL, and 5AL (Ainsworth et al. 1983) and of endosperm peroxidase on 4AL, 7AS, and 7DS (Kobrehel and Feillet 1975; Kobrehel 1978; Benito and Pérez de la Vega 1979).

Six clones (BCD 87, BCD 93, BCD 1302, CDO 780, CDO 1312, and WG 114) revealed putative interhomoeologous translocations which support the above-proposed translocations (Table 5). For 4 of these clones, 2 of the 3 major fragments were assigned to their respective homoeologous chromosomes in two of the genomes, whereas the 3rd was assigned to a different chromosomal group in the remaining genome. In concurrence with the 4AL > 5AL > 7BS > 4AL translocations proposed by Naranjo et al. (1987), we detected 4AL-specific fragments on 5AL (clones BCD 1302, CDO 1312, and WG 114), a 5AL-specific fragment on 7BS (clone BCD 87), and 7BS-specific fragments on 4AL (clones BCD 93 and

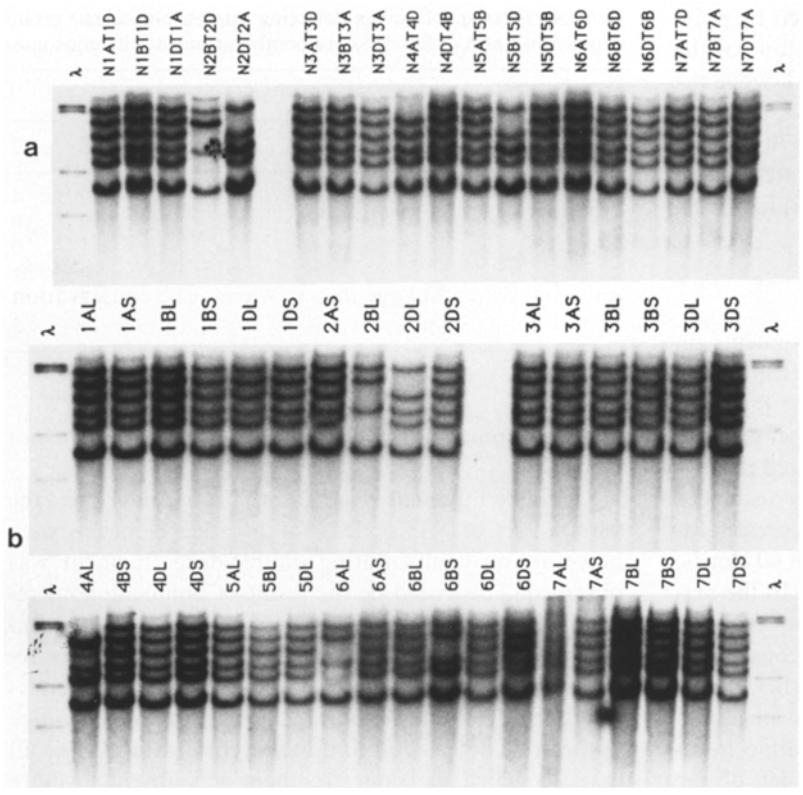

Fig. 3a, b. Evidence for the occurrence of homoeologous recombination from autoradiogram of clone BCD 348 probed onto 'Chinese Spring' wheat aneuploids digested with *Eco*RV. λ indicates lambda DNA digested with *Hin*dIII and used as a size marker. **a** Result of probing onto nullisomic(*N*)-tetrasomics(*T*). Note the concomitant absence of restriction fragments in N2DT2A and N5BT5D and the presence of double-dose fragments in N2DT2A also at higher relative intensity in N5BT5D. **b** Result of simultaneous probing onto ditelosomic stocks. Fragments assigned to group 2 chromosomes are further located to 2S based on their absence in 2BL and 2DL. Fragments absent in 6AL and 6BS may also be the result of homoeologous recombination or polymorphism

Table 5. Assignment of DNA restriction fragments to chromosome arms of clones revealing translocations

Clone designation[a]	Enzyme	Number of fragments	Fragment locations
Xcnl. BCD 87	*Dra*I	3	7BS, 5BL, 5DL
Xcnl. BCD 87	*Eco*RV	3	7BS, 5BL, 5DL
Xcnl. BCD 93	*Dra*I	6	4AL, 7AS, 7DS, 7DS[b]
Xcnl. BCD 93	*Eco*RV	7	7AS, 7AS[b], 4AL, 7DS(2)
Xcnl. BCD 1302	*Dra*I	3	5AL, 4BL, 4DL
Xcnl. CDO 484	*Eco*RV	3	4AL, 5BL, 5DL
Xcnl. CDO 484	*Eco*RI	4	4AL, 5BL, 5DL, 2DL[b]
Xcnl. CDO 780	*Eco*RI	3	4AL, 7AS, 7DS
Xcnl. CDO 780	*Dra*I	3	4AL, 7AS, 7DS
Xcnl. CDO 1312	*Dra*I	5	5AL(2), 1BL, 4BL, 4DL
Xcnl. CDO 1312	*Eco*RI	5	5AL, 1BL, 4BL, 4DL
Xcnl. WG 114	*Dra*I	3	5AL, 4BL, 4DL
Xcnl. WG 114	*Eco*RI	3	5AL, 4BL, 4DL

[a] RFLP markers are designated by an "X" followed by the institution code (cnl = Cornell University

[b] Represent minor fragments; all others represent major fragments

CDO 780). In addition, we detected a 5AL-specific fragment on 4AL (clone CDO 484) (Table 5). One possible explanation of these results that expands upon Fig. 4 of Naranjo et al. (1987) is illustrated in Fig. 4. In the A genome progenitor of hexaploid wheat a reciprocal translocation occurred between terminal segments of the long arms of chromosomes 4 and 5. In the tetraploid (AABB), the rearranged 4AL exchanged terminal segments with 7BS via a reciprocal translocation. The reasons for the occurrence of the translocations in the diploid and tetraploid, respectively, were discussed by Naranjo et al. (1987). Our proposal adds to that of Naranjo et al. (1987) in that a segment of 5AL was transferred to 4AL. This segment is most likely interstitial, thus allowing for the 4AL-7BS reciprocal translocation to involve terminal portions of the chromosomes.

Polymorphism among stocks and other anomalies

Polymorphism (here an observable change in the molecular weight of 1 or more fragment compared with 'Chinese Spring') was observed with 21 different clones on the aneuploid stocks. Ditelosomic stocks 4AL and 7BL were polymorphic for 12 and 5 clones, respectively, with other aneuploids involved in 0, 1, or 2 each. A portion of the 14 clones with fragments absent in more than one lane (excluding group 2 homoeologous recombination in N5BT5D) may fall in this category also. Clone CDO 395, located on chromosome 3S, produced more intense 3DS fragments (approximately 10×) in the DT 6BL stock than the same molecular weight fragment in other stocks. This suggests some form of localized duplication of this locus on chromosome 3DS in the DT 6BL stock.

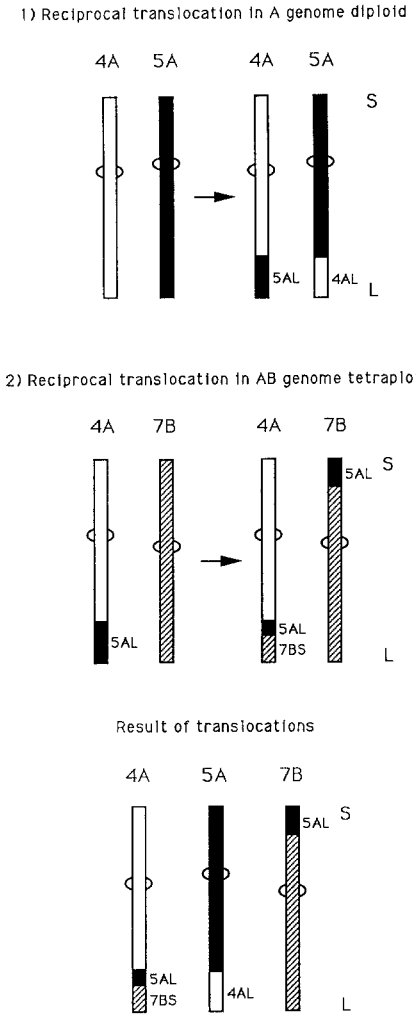

Fig. 4. Presumed sequence of translocations during the evolution of 'Chinese Spring' wheat. The approximate location of centromeres, based on somatic arm ratios, are indicated by *ovals*. The actual sizes of the translocations are not not known

Twenty-eight of the clones were mapped using more than one enzyme to confirm fragment locations when there were ambiguities. The use of more than one enzyme often resulted in a different number of fragments localized within a chromosomal group. This was expected since only clones giving ambiguous or incomplete results were probed with additional enzymes. Only 4 clones (BCD 348, CDO 484, CDO 836, and CDO 1400) yielded fragments assigned to different homoeologous groups from the use of the different enzymes. The results from BCD 348 and CDO 1400 may be largely due to polymorphism since these two clones revealed the highest levels of polymorphism compard to 46 other clones probed onto 18 hexaploid wheat genotypes (Anderson et al. in preparation).

The anomalies detected (translocations, homoeologous chromosome pairing, and polymorphism among DNAs) in these aneuploid stocks using molecular markers dictate that caution should be exercised in interpreting the results from genetic studies utilizing aneuploid stocks.

Applications of an arm-specific map

We view the development of this chromosomal arm map as a complement to, rather than a substitute for, a conventional RFLP linkage map in wheat. The conservation of gene synteny among the three genomes of wheat and relatives such as *T. tauschii* (DD) (Kam-Morgan et al. 1989; Gill et al. 1991) barley and rye (Hart 1987) means that linkage maps produced in these diploids may be suitable for use on hexaploid wheat. Linkage maps should be more efficienctly produced using diploid relatives of wheat such as *T. tauschii* (Kam-Morgan et al. 1989; Gill et al. 1991); *T. monococcum* (A genome) (M. Röder personal communication) or barley (Heun et al. 1991) since higher levels of polymorphism can be found and only a single genome needs to be mapped.

The efficiency and usefulness of a chromosomal arm map versus a linkage map in wheat will vary depending on the application. Since polymorphisms in cultivated bread wheat are relatively rare for any given pair of lines (Chao et al. 1989; Kam-Morgan et al. 1989; Liu and Tsunewaki 1990; unpublished data), a large pool of clones will be required for selecting those informative on specific populations. The initial construction of a chromosomal arm map is relatively rapid since all low-copy clones that are polymorphic among the A, B, and D genomes can be mapped using aneuploids. A disadvantage of conventional RFLP linkage mapping is that only those clones polymorphic for at least 1 fragment are mapped on the specific mapping population, thus eliminating clones that may be informative in other populations (Anderson et al. in preparation). In addition, linkage mapping populations are often constructed from a cross of distantly related genotypes within the primary gene pool or from different gene pools. The more distantly related the parents, the more polymorphism can be expected, but there may be a greater risk of cytological abnormalities such as reduced recombination and the presence of translocations or other chromosome rearrangements. This was encountered by Chao et al. (1989) for chromosome 7D of wheat in a cross in which one parent had an alien chromosome segment.

A chromosomal arm map should be especially useful in deciphering genetic relatedness of varieties and species accessions based on RFLPs since one could choose clones that represent all chromosome arms. Clones from this arm map may also find immediate application in the field of wheat cytogenetics as previous outlined (Gale et al. 1989). A subset of clones may be useful in following the introgression of alien chromosome segments, reduc-

ing or eliminating linkage drag (Young and Tanksley 1989), detecting changes in cytogenetics stocks, constructing addition or substitution lines, and detecting other cytological abnormalities.

The tagging of qualitatively inherited traits should be enhanced by knowing the chromosomal arm location of clones. Not only does this make the search for unmapped genes more efficient, but in the case of wheat many genes of economic importance have already been located to chromosome arms of particular genomes (Milne and McIntosh 1990; Hart and Gale 1990). Putative linkages to genes of economic importance have been identified in our laboratory using clones from the chromosomal arm map. These include linkages to genes for resistance to Hessian fly (*Mayetiola destructor*) (Z Ma, personal communication), leaf rust (*Puccinia recondita* f. sp. *tritici*), and stem rust (*Puccinia graminis* f. sp. *tritici*) (E Autrique, personal communication).

The identification of quantitative trait loci (QTL) is best facilitated by a linkage map because of the need for uniform genome coverage to ensure detection of as many QTLs as possible (Paterson et al. 1988). This can be accomplished in the case of wheat by selecting clones from linkage maps of related diploids and supplementing those with clones on the arm map in order to adequately cover the chromosomes of the hexaploid with polymorphic markers.

One of the drawbacks of a chromosomal arm map is that the linear arrangement and genetic linkage of clones is not known. Knowing the linear arrangement of clones would make screening for linkages with genes of interest more efficient. However, polymorphic clones must be mapped in the population segregating for the trait of interest regardless of whether they come from an arm map or a linkage map. A second drawback of a chromosomal arm map is that the clones have been selected for intergenomic versus intragenomic polymorphism. As a result, the clones should be most useful for introgression studies, but only a subset will be informative (with today's technology) in crosses between cultivated wheat varieties.

The complementation of RFLP mapping with a chromosomal map is likely to be efficient for other species in which the level of polymorphism is low, and aneuploid stocks are available that allow the placement of clones to chromosome arms.

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